

AGRAULIS AND PASSIFLORA II. BEHAVIOR AND SENSORY MODALITIES¹

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In a previous paper (Copp and Davenport, 1978) we described for the heliconian *Agraulis vanillae incarnata* (Riley) experiments on larval feeding preferences, adult ovipositional preferences, food-plant suitability of various species of *Passiflora*, and a field survey of passion-vine infestation in Santa Barbara County, California. The results indicated that, more than any other factor investigated, ovipositional preferences determine the infestation levels of the various local species of *Passiflora*. All remarks to follow concerning these aspects of the *Agraulis-Passiflora* relationship refer to the paper cited above.

If the interface of most direct interaction between the two organisms is the selection of specific passion-vines as larval food, it follows that the sensory modalities and consequent behavior governing this selection can be expected to have important effects on the evolution of both butterfly and plant. The following experiments were designed to investigate the modalities and behavior. Results are discussed in the light of the above-cited field studies.

MATERIALS AND METHODS

The butterflies and plants

All procedures for raising *Agraulis* and collecting *Passiflora* plant material have been discussed in the preceding paper. "Host-plant" was defined as that species of *Passiflora* on which the adult butterflies had been raised as larvae. *Passiflora alato-caerulea* is hereafter referred to as AC; *P. caerulea* as CR; *P. manicata* as MN; *Passiflora* sp. as SP; *P. mollissima* as ML; and *P. edulis* as ED.

Visual selection of ovipositional sites

It has been suggested that *Heliconius* females select species of *Passiflora* for oviposition on the basis of leaf shape (Gilbert, 1975). The possibility that specific visual cues might function in the ovipositional preferences of *Agraulis* was tested in the following manner.

A single new-growth shoot of a passion-vine (ca. 5 leaves with the cut end in 10 ml water) was placed inside an inverted one-gallon glass container fitted with an air-tight lid. Four, 2.5 cm-wide strips of cheesecloth, taped to the top and sides of the container, facilitated landing and oviposition by the female butterflies.

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This was placed in a cage (61 cm square, 90 cm tall) beside an identical container either left empty or containing a new-growth shoot from a different species of plant. In experiments involving two species of vine, the appropriate removal of leaves equalized the surface area presented by each.

An experimental trial consisted of placing two containers, comprising an experimental pairing, in a cage with females (four days old, mated, and raised as larvae on either SP or CR), reversing their position after 24 hours, and counting the number of eggs on each after 48 hours. Each experimental pairing was conducted three times under laboratory culture conditions (23° C, 60% humidity; 15L:9D).

Three different plant species were used in these experiments: AC, the vine most oviposited upon by *Agraulis* in laboratory preference tests; ED, the vine least oviposited upon in ovipositional preference tests; and *Hedera canariensis* (Algerian Ivy), a readily available plant not oviposited upon by *Agraulis* in the laboratory. Leaves of these three plants differ sufficiently in both color and shape to make them easily distinguishable by the human observer. In a fourth experimental pairing a container with leaf models, cut from green construction paper to the shape of AC leaves, was paired with an empty control container.

The butterflies had had no previous exposure to plant material prior to all experiments except those matching AC with *Hedera*. In the excepted case, four-day old females were allowed to oviposit on sprigs of AC and *Hedera* in the laboratory for an additional four days immediately preceding the test. The increased age of the females in this test probably had no effect on the results since oviposition behavior has not been observed to change during the period of active oviposition in a female's life span.

Volatile stimulants of oviposition

Observations of females in the laboratory suggested that a volatile agent, emitted by some species of *Passiflora*, accelerated oviposition by *Agraulis* females. The following experiment examines the effect on ovipositional behavior of substances volatilized from *Passiflora* leaves.

A three-gallon glass container with an air-tight lid was equipped with a 500 ml Nalgene bottle and a wire screen cylinder such that females placed in the glass container were able to "smell" vine enclosed in the Nalgene bottle but neither make contact with it nor see it. About 150 holes, each 1.5 mm in diameter, were drilled in the side of the Nalgene bottle, and its lid was attached with white glue to the inside of the lid of the glass container. A wire screen cylinder (10.5 cm in diameter, 21 cm tall), closed at the bottom, fitted into the neck of the glass container and prevented tarsal and proboscis contact by the butterfly with the vine contained inside the Nalgene bottle. Odors could circulate freely.

Prior to an experiment, ten mated, four-day old females were isolated from their cages and fed a 1 M sucrose solution. Five females were then placed in each of two glass containers (one experimental, one control), the wire cylinders inserted, and the containers' lids screwed down, thus suspending the Nalgene bottles within the wire cylinders. Ten 20 cm long sprigs of fresh new-growth shoots of *Passiflora* (50 leaves), placed with water in the Nalgene bottle, provided the olfactory

stimulus. Females oviposited primarily on a circle of filter paper placed on the floor of the glass container. Experimental containers with vine and control containers without vine were placed under the cage lights in the culture laboratory, left 24 hours, reversed, and left another 24 hours. The number of eggs in each was counted after 48 hours. Each experimental situation was replicated six times involving a total of 60 females.

Washing all pieces of equipment with 50% ethanol after each experimental replicate and randomizing their assembly helped to control for residual, adsorbed odors. Presumably, humidity levels did not differ between paired bottles. Thermometers, taped to the inside of the glass containers, indicated that in all cases both experimental and control containers became equally warm over the course of the experiment (30° C).

Anemotactic behavior

The ease with which a potential host-plant is located by gravid *Agraulis* females will partially determine that vine's level of infestation, assuming that females spend an appreciable amount of time searching for ovipositional sites. It has long been known that some adult lepidopterans exhibit a positive anemotaxis (directed movement upwind) when stimulated by certain olfactory agents such as sex pheromones. The possibility that *Passiflora* odors elicit a positive anemotaxis from *Agraulis* females was tested in the following experiments.

A four- to six-day old female was tethered in the following manner to allow restricted flight in any direction: one end of a length of light weight thread was attached to the butterfly's thorax with a small piece of bubble-gum (a biologically inert adhesive with immediate "stickiness," low price, and recreational value) and the other end passed through a 15 cm length of small diameter glass tubing clamped vertically to a ring stand. The butterfly's thorax was drawn up within 5 cm of the glass tube allowing that much forward flight and unrestricted turning.

The tethered insect could then be subjected to a slow airstream (ca. 0.9 m/sec) created by an electric fan, the speed of which was regulated by a Powerstat variable autotransformer. The fan, mounted 0.66 meters from the butterfly, directed air over a group of plant shoots (about 200 leaves) placed beneath the butterfly's visual field. A square card, divided into four equal quadrants, was taped to the table directly beneath the suspended butterfly. A line bisecting one of the quadrants pointed towards the center of the fan. Forty-five degrees either side of this line was defined as the "upwind" direction. The orientation of a butterfly in stationary flight could then be monitored by noting, from above, the quadrant within which the extended tether-string fell. This apparatus was placed in a small room separate from the culture laboratory but under the same temperature and humidity conditions (23° C, 60% humidity).

Twenty-four hours prior to an experiment the females to be tested were selected from their cages, with no preference for host-plant, and placed without vine in a holding cage in the separate test laboratory. Just before bringing vine into this room all females were further isolated in air-tight, one-gallon glass containers, five females per container, to minimize their exposure to vine odors prior to testing. A female, selected from one of the containers and attached to the tether, stood on a small

platform for one minute. The fan was turned on at the end of this period and the platform removed. A single trial began with the consequent, reflex onset of flight. The butterfly was randomly oriented with respect to the direction of the airstream at that time. Each trial lasted for five minutes during which the amount of time that the tether pointed in the upwind direction was recorded with a stopwatch. All experiments began at 10:00 AM, four hours after the butterfly's "dawn." Thirty females, in blocks of ten, comprised three replicates of both experimental and control situations. Only those females that flew the entire five minutes were recorded, and each flew only once.

Specimens of AC, MN, and ML were used as odor sources because they represent high, median and low points in the spectrum of ovipositional preferences and in observed levels of infestation in the field. Control experiments omitted the vine or substituted *Hedera canariensis* for *Passiflora*.

Because the laboratory became contaminated with plant odors during the experimental trials, it was not possible to subject one butterfly to a control trial immediately followed by an experimental trial on the same butterfly and still accumulate an adequate sample size in a reasonable amount of time. Control and experimental trials occurred several days apart and involved different groups of equally old females.

Dispersal of adult Agraulis with respect to host-plants

The relative significance of visual cues, volatile stimulants and inhibitors of oviposition, and olfactory orientation to host-plants in determining the level of infestation of various species of *Passiflora* by *Agraulis* will vary with the degree of active host seeking and dispersal of the ovipositing females. The movements of adult butterflies among several separate passion-vines were observed during a mark-release-recapture study conducted in Isla Vista, California in the summer of 1975.

All captures of butterflies occurred at one of three passion-vines which formed a triangle (site I to site II, 385 m; I-III, 220 m; II-III, 325 m). No other passion-vines existed within or near this triangle to our knowledge. Prevailing wind was from WSW (*i.e.*, approximately) from site I (MN vine) toward sites II and III (AC vines). An observer stood near a vine and captured as many *Agraulis* butterflies as possible within a one meter radius of the vine for a period of one hour in the late morning or early afternoon. Butterflies were individually marked by writing a number on the underside of the wings with a "Sharpie" laundry marker. Numbering was easy, permanent and permitted recording of an individual's movements among the three vines. When released, the numbered butterflies flew away immediately and showed no ill effects as a result of the handling. The number, sex, and time of capture for each butterfly was recorded according to location.

Each of the three locations was sampled every day for the first two weeks beginning July 10, 1975 and three times a week thereafter until October 13, 1975. The three vines were always sampled consecutively on the same day. The order of their sampling was random. At each sampling the observer recorded the marks of all recaptured butterflies (those numbered prior to the sampling). Multiple

TABLE I

Results of tests for visual discrimination of different species of plants by ovipositing *Agraulis*. Each experimental trial lasted 48 hours. *P* was calculated by Student's *t*-test for paired comparisons ($P > 0.05$; *n.s.*, not significant). *AC* represents *Passiflora alato-caerulea*; *ED*, *P. edulis*; and *Hedera*, *Hedera canariensis*.

Experimental pairing	Trial number	Number of females	Number of eggs	<i>P</i>
AC/blank	1	32	97/ 16	0.01 < <i>P</i> < 0.02
	2	10	59/ 19	
	3	17	110/ 38	
	total	59	266/ 73	
AC/ED	1	31	47/ 15	<i>n.s.</i>
	2	31	23/ 26	
	3	39	102/133	
	total	101	172/174	
AC/ <i>Hedera</i>	1	30	70/ 44	<i>n.s.</i>
	2	31	35/ 43	
	3	25	25/ 88	
	total	86	130/175	
leaf model/blank	1	20	77/ 18	0.002 < <i>P</i> < 0.01
	2	10	52/ 3	
	3	10	59/ 1	
	total	40	188/ 22	

recaptures of a single butterfly in one day were recorded for each event, but only the first recapture on that day entered into the calculation of movement patterns unless they occurred at different sites.

RESULTS

Visual selection of ovipositional sites

The results of the visual selection experiments are presented in Table I. The container with AC vine and the one with the green paper leaf model were both more oviposited upon than the empty control containers. The AC container did not receive significantly more eggs than either the ED or the *Hedera* container.

Prior to the third experimental pairing, females had been allowed to oviposit on fresh sprigs of AC and *Hedera*; during this time the latter plant received no eggs, while AC was heavily oviposited upon. After this 4-day period of freedom of access the same females were given the same choice in the absence of chemical cues; under such circumstances they made no significant choice. Apparently, as the results of this test show, the females did not, in the time available, learn to associate leaf color and shape with a suitable ovipositional stimulus.

TABLE II

Effects of Passiflora odors on oviposition by Agraulis. The number of eggs is the total number laid by thirty females during six replicate experiments. In each replicate, a group of ten females was divided equally between two containers; one with Passiflora, and one without (blank). P was determined by Student's t-test for paired comparisons ($P > 0.05$; n.s., not significant). AC represents P. alata-caerulea; CR, P. caerulea; MN, P. manicata; SP, P. sp.; ML, P. mollissima; and ED, P. edulis.

Experimental pairing	Number of eggs	P
AC	130	$P < 0.05$
Blank	37	
CR	955	$P < 0.05$
Blank	570	
MN	314	$P < 0.05$
Blank	464	
SP	308	$P < 0.05$
Blank	506	
ML	381	n.s.
Blank	372	
ED	116	n.s.
Blank	129	

In preliminary experiments (Copp, 1976) which monitored oviposition when passion-vine (AC) either was or was not visible to the females, two additional points pertinent to the role of visual cues in oviposition were revealed: first, when direct contact was prevented, eggs were laid adjacent to passion-vine only when the vine could be seen; and secondly, maximal rates of oviposition occurred only when females had full access (visual, olfactory, and tactile) to the vine.

All of these results indicate that ovipositing *Agraulis* females preferentially localized their eggs on containers with green, leaf-shaped objects but did not visually prefer one species of *Passiflora* over another, nor did they visually select a *Passiflora* over the nonpassifloraceous plant, *Hedera*. Large numbers of eggs were even deposited on a container with crude paper leaf models inside. Apparently, visual cues are important as generalized attractants of ovipositing females rather than as indicators of specific host-plant species.

Volatile stimulants of oviposition

Although the replicates of each experimental situation occurred within two weeks of each other and involved females of similar history, different situations were tested at different times of the year with females raised as larvae on different species of *Passiflora*. This prevents extensive comparison between experimental situations. However, the results (Table II) indicate that: AC and CR emitted some factor which significantly increased the rate of oviposition in those containers over the controls; ML and ED odors had no effect on the rate of oviposition; and odors emitted by MN and SP inhibited oviposition slightly. The unexpectedly high rates of oviposition in most of the control containers may

TABLE III

Amount of time out of five minutes that tethered *Agraulis* females faced upwind in airstreams labeled with various plant odors. Mean time represents the mean upwind flight time for thirty females. *P* was calculated by Student's *t*-test comparing the mean with a hypothetical value of $\mu_0 = 75$ sec ($P > 0.05$; n.s., not significant). The standard error of the mean is given in seconds. AC represents *Passiflora alato-caerulea*; MN, *P. manicata*; ML, *P. mollissima*; and Hedera, *Hedera canariensis*.

Airstream treatment	Mean upwind flight time/5 min (min:secs)	<i>P</i>
Unlabeled	1:00 \pm 6.2	$P > 0.05$
AC	2:23 \pm 11.5	$P < 0.001$
MN	2:21 \pm 16.2	$P < 0.001$
ML	1:12 \pm 10.0	n.s.
Hedera	1:13 \pm 10.4	n.s.

have resulted from inadvertent selection for spontaneous oviposition in laboratory cultures of insects as previously discussed (Copp and Davenport, 1978).

In the ovipositional preference tests the mean number of eggs laid per female was highest in those experiments which included either AC or CR as one of the choices. This phenomenon may now be explained by the presence in those tests of volatile stimulants of oviposition released by AC and CR.

The effects of volatile compounds, as noted in these experiments, cannot account entirely for the degree of resolution of host-plant species evident in the ovipositional preference experiments. Specimens of MN and SP for example, were consistently more oviposited upon than either ML or ED, yet the former two species emitted a volatile inhibitor of oviposition not present in ML and ED leaves. Also, as mentioned above, maximal rates of oviposition occurred only when the females were free to contact the vine. It seems likely that contact-chemical cues, in addition to volatile chemical signals, function in determining ovipositional preferences. This hypothesis is supported by observations that female specimens of *Agraulis* frequently "drum" a leaf's surface with their foretarsi just before ovipositing. Drumming is a behavioral act thought to allow "tasting" of a leaf's chemical constituents by the female butterfly (Ilse, 1937; Fox, 1966; Ma and Schoonhoven, 1973). Numerous attempts were made to develop an effective bioassay for the detection of these presumed contact-chemical cues in *Passiflora* leaves, but with no conclusive success to date. The chromatographic "spot" patterns of extracts from fresh *Passiflora* leaves have been compared. No correlations with ovipositional preference have been observed. Differences in leaf texture among the six species of *Passiflora* studied appeared minimal and generalized, and were not considered to have a significant influence on specific ovipositional preferences.

Anemotactic behavior

For a female butterfly which is orienting randomly with respect to the direction of the airstream, the upwind flight time should not differ significantly from 1 minute and 15 seconds, *i.e.*, one quarter of the trial period. Significantly non-

TABLE IV

Mark-release-recapture data for *Agraulis* in Isla Vista, California, July 10 to October 13, 1975. Site I is a MN vine; sites II and III are AC vines. The "mark site" is the site at which the butterfly was originally marked.

Site	Number of butterflies marked	Number recaptured at least once	Total number of recaptures	Number recaptures	
				At mark site	At other sites
I	203	127	337	296 (87.8%)	41
II	120	82	188	180 (95.7%)	8
III	39	17	36	29 (80.6%)	7

random upwind orientation by *Agraulis* females occurred only when the airstream had passed over either AC or MN vine (Table III). Females did not face preferentially upwind into an unlabeled airstream or ones which had passed over either *Hedera canariensis* or ML vine. Indeed, females exhibited a slight tendency to face down- or across-wind in an unlabeled airstream. Variations in the age of females or their sensitivity to plant odors may have caused the high variability in the experiments involving AC and MN.

Although olfactory cues have been shown to be involved in the localization of host-plants by insects in other orders (e.g., Moorehouse, 1971; Werner, 1972), this may be the first experimental demonstration of such a capability in a lepidopteran. The results of Yamamoto and Fraenkel (1960), Schurr and Holdaway (1970), and Wearing, Connor and Ambler (1973), which may indicate chemically triggered orientation by lepidopterans, are open to other interpretations.

Dispersal of adult butterflies with respect to host-plants

Monitoring the movements of *Agraulis* among three passion-vines revealed that the adults remain highly localized with respect to larval food resources (Table IV). Despite observations that *Agraulis* is a strong flier, lives up to four weeks in the field, and forages widely for nectar, there was little movement of adults among vines 220 to 385 meters apart.

The problem of correcting for immigration, birth, and death prevented accurate estimates of population size. Hereafter the apparent size of a subpopulation of *Agraulis* refers to the number of butterflies captured at a particular site over the four month period.

To test whether the butterflies that were repeatedly recaptured at any one of the three vines sampled consisted of adults which had emerged on or near that vine, 40 pupae (36 at site II, 4 at site I) were enclosed in loose cheesecloth sacks and the adults marked upon emergence. All but eight died prior to marking, mostly as a result of tachinid fly parasitization, but also as a result of incomplete emergence. The recapture records of the few surviving adults support the contention that adults return repeatedly to their larval food plant's site. Of the seven adults that emerged at site II, six were recaptured a total of twelve times, each time at site II. One adult from site II was never recaptured. The one adult that emerged at site I was recaptured three times only at site I.

A more detailed look at the instances of emigration between sites reveals the following points: first, of the 41 butterflies that moved between vines at least once, 24 were female and 17 male. Males and females exhibited no significant difference in their tendencies to emigrate ($P < 0.05$, chi-squared test for goodness-of-fit); secondly, the apparent size of a site's subpopulation did not correlate with the proportion of marked butterflies that emigrated from that site as was found to be true for *Pieris* (Shapiro, 1970). Site I, a large MN vine with an apparently dense subpopulation, yielded 30 of the 41 emigrants, *i.e.*, 15.2% of the total number of butterflies marked at site I moved between sites at least once. This compares with 12.8% emigrants from site III, a small, relatively sparsely populated, AC vine, and 5.0% from site II, an AC vine of intermediate size and population density; and thirdly, emigrants did not favor any site for arrival. The 30 emigrant adults numbered at site I made 13 trips to site II, 19 to site III, and only eight return trips. The six emigrants from site II made a total of six trips, three to each of the other sites, and no return trips. Three of the five emigrants from site III went to I, two to site II and none made return trips. Considering the results of the adult flight orientation experiments and the prevailing westerly wind direction, emigrating butterflies had been expected to move upwind toward site I, but they did not do so.

DISCUSSION

The foregoing experiments indicate that the ovipositional preferences of *Agraulis*, as exhibited in the laboratory, can be explained as graded responses to various ratios of volatile and contact-chemical signals which affect oviposition. The ovipositional responses may be triggered (*e.g.*, AC, CR, MN, SP) or inhibited (*e.g.*, ML and ED) by contact-chemical stimuli. The rate of oviposition after triggering could be partially determined by volatile stimulants (*e.g.*, AC and CR) or inhibitors (*e.g.*, MN and SP). Gradations in quantity or differences in quality of either chemical component could account for finer resolution of host-plant species by ovipositing females. Pre-ovipositional behavior and oviposition in several species of Lepidoptera have been shown to be affected by both volatile cues (*e.g.*, Vaidya, 1969; Städler, 1974; Sutherland, Hutchins and Wearing, 1974) and contact-chemical cues (*e.g.*, Yamamoto, Jenkins, and McClusky, 1969; Ma and Schoonhoven, 1973).

The generalized role of vision in the selection of ovipositional sites by *Agraulis* is not surprising in view of similar findings for other lepidopterans (*e.g.*, Ilse, 1937; Vaidya, 1969; Wiklund, 1974). However, it has been proposed that *Heliconius* butterflies select species of *Passiflora* for oviposition primarily on the basis of learned or innate visual responses to specific leaf shapes (Gilbert, 1975). Our experiments gave no indication of this capability in *Agraulis*.

Considering the low frequency with which *Agraulis* butterflies move between specimens of *Passiflora*, the upwind flight response does not appear to be significant in affecting host-plant selection in the field. There are other possible functions for this orientational response: it may aid in the localization of new host-plants if, in response to extreme host-plant depletion, *Agraulis* dispersal increases. Since *Agraulis* larvae frequently pupate some distance from the host-plant,

the upwind flight response may serve in the initial localization of the host-plant by the newly emergent adult. A third possible function is that it may, in effect, "trap" adults around their particular larval food plant. None of these possibilities excludes the others, and each may work in concert with visual orientation to learned landmarks. The ability of other adult heliconians to return repeatedly to a single site is evident in their "trapping" of pollen resources (Ehrlich and Gilbert, 1973).

The division of a population of butterflies into a number of subpopulations, each centered on a different patch of plant resources, has been observed in *Heliconius* (Ehrlich and Gilbert, 1973) and *Euphydryas editha* (Ehrlich, 1961; Gilbert and Singer, 1973; Ehrlich and Singer, 1974). For these butterflies adult dispersal seems to depend mainly on the distribution of the adult's food resources. In contrast, subpopulations of *Agraulis* in Santa Barbara County were found to be centered on single larval food-plants. *Agraulis* in the tropical zone might be expected to disperse more widely among larval food plants as a consequence of increased parasitic pressure and competition for food, much as *Heliconius* avoids local concentrations of parasites or larval overcrowding by spreading out its eggs in both space and time (Gilbert, 1975).

In southern California it would seem that the pattern of *Passiflora* infestation does not result from active choice by female butterflies searching for ovipositional sites, as much as from the "trapping" of sedentary populations on specific plants, the levels of infestation depending primarily on the balance of compounds affecting oviposition particular to that species of *Passiflora*. The following model for local host-plant infestation by *Agraulis* is proposed. A previously uninfested passion-vine, or one which has lost its population by winter-killing, is located by a female, possibly as a consequence of an upwind flight response to host specific odors followed by a series of nonspecific, visually directed test landings. Oviposition is apparently triggered or inhibited by appropriate contact-chemical cues. If triggered, it proceeds at a rate dependent on the concentration of chemical stimulators and/or inhibitors (volatile and nonvolatile). Following successful development, subsequent generations of females oviposit on the same individual plant. The newly established subpopulation of butterflies increases at a rate which depends primarily on the rate of oviposition and, secondarily, on the rate of larval and pupal mortality on that species of vine.

The local association between *Agraulis* and *Passiflora* could be severely affected at any number of control points at each step of this proposed model. These control points need not necessarily involve plant chemistry. For example, Gilbert (1971) has demonstrated that hooked trichomes on the leaves of *Passiflora adenopoda* act as a highly effective barrier to heliconian infestation by virtue of their lethal effect on the larvae. Also, Singer (1971) has demonstrated in *Euphydryas editha* that the growth forms and microhabitats of various potential host-plant species determine which is most infested in a single locale by a particular population of the butterfly. However, the *Agraulis*-*Passiflora* association in Santa Barbara County appears to hinge on plant chemistry, more particularly, on these chemical factors directly involved in the butterflies' specific ovipositional responses. Plant chemistry has been shown to be an important element in the coevolutionary rela-

tionships between other lepidopterans and their host-plants (Ehrlich and Raven, 1964; Dolinger, Ehrlich, Fitch and Breedlove, 1973).

It would broaden our understanding of the coevolutionary relationship between heliconians and passion-vines to examine further the role of plant chemistry in host-plant localization and oviposition by the butterflies and to conduct an in-depth chemical survey of the family Passifloraceae from the standpoint of the chemical control of butterfly behavior.

We would like to thank Mr. Stephen Maskel whose accuracy with a butterfly net helped us considerably in the mark-release-recapture study.

SUMMARY

1. The behaviors and sensory modalities involved in the selection of *Passiflora* host-plants for oviposition by the heliconian butterfly *Agraulis vanillae* were investigated.

2. Ovipositing females are attracted to oviposit near green leaf-shaped objects on the basis of generalized visual cues but make no ovipositional distinction between *P. alato-caerulea* and *P. edulis*, which are readily discriminated when full access to the vines is allowed.

3. The rate of oviposition by female *Agraulis* is accelerated by odors from *P. alato-caerulea* and *P. caerulea*, retarded by odors from *P. manicata* and *Passiflora* sp., and not affected by odors from *P. edulis* or *P. mollissima*.

4. Contact-chemical stimulants and inhibitors of oviposition are presumed to exist in the leaves of passion-vines on the basis of circumstantial evidence. Attempts to demonstrate directly their existence have not yet been successful.

5. Female *Agraulis* exhibit a positive anemotactic response in an airstream that has passed over one of at least two species of *Passiflora*, *P. alato-caerulea* and *P. manicata*, but not in an airstream passed over *P. mollissima*.

6. A mark-release-recapture study of adult movements among three passion-vines demonstrated that butterfly subpopulations tend to form on each vine with little exchange between different subpopulations.

7. The primary factor determining the infestation level of a particular *Passiflora* by *Agraulis* is the balance of chemical cues directly involved in oviposition that are present in that *Passiflora*. Possible functions for the upwind flight response are suggested. Further research on the plant chemistry of the family Passifloraceae as it relates to heliconian behavior would broaden our understanding of heliconian-*Passiflora* coevolution.

LITERATURE CITED

- COPP, N. H., 1976. Host-plant selection by the Gulf fritillary butterfly *Agraulis vanillae incarnata* (Riley). *Ph.D. Dissertation, University of California, Santa Barbara*, 117 pp.
- COPP, N. H., AND D. DAVENPORT, 1978. *Agraulis* and *Passiflora*. I. Control of specificity. *Biol. Bull.*, **155**: 98-112.
- DOLINGER, P. M., P. R. EHRLICH, W. C. FITCH, AND D. F. BREEDLOVE, 1973. Alkaloid and predation patterns in Colorado lupine populations. *Oecologia*, **13**: 191-204.

- EHRlich, P. R., 1961. Intrinsic barriers to dispersal in a checkerspot butterfly. *Science*, **134**: 108-109.
- EHRlich, P. R., AND L. E. GILBERT, 1973. Population structure and dynamics of the tropical butterfly *Heliconius cthylla*. *Biotropica*, **5**: 69-82.
- EHRlich, P. R., AND P. H. RAVEN, 1964. Butterflies and plants; a study in coevolution. *Evolution*, **18**: 586-608.
- EHRlich, P. R., AND M. C. SINGER, 1974. Adult movements and population structure in *Euphydryas citha*. *Evolution*, **28**: 408-415.
- FOX, R. M., 1966. Forelegs of butterflies I. Introduction: chemoreception. *J. Res. Lepid.*, **5**: 1-12.
- GILBERT, L. E., 1971. Butterfly-plant coevolution: has *Passiflora adenopoda* won the selectional race with heliconiine butterflies? *Science*, **172**: 585-586.
- GILBERT, L. E., 1975. Ecological consequences of a coevolved mutualism between butterflies and plants. Pages 210-240 in L. E. Gilbert and P. H. Raven, Eds., *Coevolution of animals and plants*. University of Texas Press, Austin.
- GILBERT, L. E., AND M. C. SINGER, 1973. Dispersal and gene flow in a butterfly species. *Am. Nat.*, **107**: 58-72.
- ILSE, D., 1937. New observations on responses to colours in egg laying butterflies. *Nature*, **140**: 544.
- MA, W. C., AND L. M. SCHOONHOVEN, 1973. Tarsal contact chemosensory hairs on the large white butterfly *Pieris brassicae* and their possible role in oviposition behavior. *Entomol. Exp. Appl.*, **16**: 343-357.
- MOOREHOUSE, J., 1971. Experimental analysis of the locomotor behavior of *Schistocerca gregaria* induced by odor. *J. Insect Physiol.*, **17**: 913-920.
- SCHURR, K., AND F. G. HOLDAWAY, 1970. Olfactory responses of female *Ostrinia nubilalis* (Lepidoptera: Pyraustinae). *Entomol. Exp. Appl.*, **13**: 455-461.
- SHAPIRO, A. M., 1970. The role of sexual behavior in density-related dispersal of pierid butterflies. *Am. Nat.*, **104**: 367-372.
- SINGER, M. C., 1971. Evolution of food plant preference in the butterfly *Euphydryas citha*. *Evolution*, **25**: 383-389.
- STÄDLER, E., 1974. Host plant stimuli affecting oviposition behavior of the eastern spruce budworm. *Entomol. Exp. Appl.*, **17**: 176-188.
- SUTHERLAND, O. R. W., R. F. HUTCHINS, AND C. H. WEARING, 1974. The role of the hydrocarbon α -farnesene in the behavior of the codling moth larvae and adults. Pages 249-263 in L. Barton Browne, Ed., *Experimental analysis of insect behavior*. Springer-Verlag, New York.
- VAIDYA, V. G., 1969. Investigations on the role of visual stimuli in the egg laying and resting behavior of *Papilio demoleus* L. (Papilionidae, Lepidoptera). *Anim. Behav.*, **17**: 350-356.
- WEARING, C. H., P. J. CONNOR, AND K. D. AMBLER, 1973. Olfactory stimuli of oviposition and flight activity of the codling moth, *Laspeyresia pomonella*, using apples in an automated olfactometer. *N. Z. J. Sci.*, **16**: 697-710.
- WERNER, R. A., 1972. Aggregation behavior of the beetle *Ips grandicollis* in response to host-produced attractants. *J. Insect Physiol.*, **18**: 423-437.
- WIKLUND, C., 1974. Oviposition preference in *Papilio machaon* L. in relation to the host plants of the larvae. *Entomol. Exp. Appl.*, **17**: 189-198.
- YAMAMOTO, R. T., AND G. FRAENKEL, 1960. The physiological basis for the selection of plants for egg laying in the tobacco hornworm, *Protoparce sexta* (Johan.). *Proc. Int. Congr. Entomol. 11th Vienna*, **3**: 127-133.
- YAMAMOTO, R. T., R. Y. JENKINS, AND R. K. McCLUSKY, 1969. Factors determining the selection of plants for oviposition by the tobacco hornworm, *Manduca sexta*. *Entomol. Ent. Appl.*, **12**: 504-508.